Citation:

Kralova Lesna I, Suchanek P, Kovar J, Stavek P, Poledne R. Replacement of dietary saturated FAs by PUFAs in diet and reverse cholesterol transport. *J Lipid Res.* 2008 Nov; 49(11): 2,414-2,418. Epub 2008 Jul 9.

PubMed ID: 18614815

Study Design:

Randomized Crossover Trial

Class:

A - Click here for explanation of classification scheme.

Research Design and Implementation Rating:

POSITIVE: See Research Design and Implementation Criteria Checklist below.

Research Purpose:

To determine if the decrease of HDL-C in a diet enriched with polyunsaturated fat is detrimental with respect to reverse cholesterol transport (RTC) by measuring change in cholesterol efflux (CHE).

Inclusion Criteria:

- Self-motivation to participate in the study
- 18 to 55 years old
- BMI less than 30kg/m^2
- Normal concentration of lipoproteins.

Exclusion Criteria:

- Recent adherence to any diet restriction before the study
- Use of any medication that could affect study outcomes
- Diabetes mellitus
- Any major illness assessed by medical history, physical examination and laboratory screening.

Description of Study Protocol:

Design

- Randomized crossover design
- Two four-week diet interventions carried out in succession without a washout period.

Dietary Intake/Dietary Assessment Methodology

- Diet composition determined by a registered dietitian (RD)
- Diet adherence monitored by dietary records and repeated sessions with a dietitian.

Intervention

- Two diets:
  - High in saturated fatty acids (SFA diet)
    - 52% SFAs
    - 34% monounsaturated fatty acids (MUFAs)
    - 14% PUFAs
  - High in polyunsaturated fatty acids (PUFA diet)
    - 26% SFAs
    - 33% MUFAs
    - 41% PUFAs
- Both diets were isocaloric (45% carbohydrate, 15% protein, 40% fat)
- Participants were asked to:
  - Not consume food other than what was provided by the study
  - Maintain usual physical activities
  - Record events that could affect the outcome of the study (like illness).

Statistical Analysis

- All results expressed as mean ±SD
- The differences between SFA and PUFA dietary periods were evaluated using a paired T-test
- The relationship between CHE and lipoprotein parameters and change of CHE and changes of lipoprotein parameters were analyzed by simple linear regression.

Data Collection Summary:

Timing of Measurements

Biochemical: Blood samples were taken after a 12-hour overnight fast at the beginning of study and end of each four-week diet period.

Dependent Variables

- Plasma triglyceride concentrations, total cholesterol and LDL-C concentrations determined enzymatically by commercial kits (Roche Diagnostics)
- HDL-C concentrations measured by phosphotungstate precipitation of apoB-containing lipoproteins
- Non-esterified fatty acid (NFEA) concentrations measured by an enzymatic test (Wako Chemicals GmbH, Neuss, Germany)
- apoA-I and apoB concentrations measured by imunoturbidimetric assay (Orion Diagnostica, Espoo, Finland)
- Anthropometric data (weight and waist circumferences) determined at the beginning of the study and at the end of the each four-week diet
- RCT (reverse cholesterol transport) measured using in vitro in cells pre-labeled in the medium containing labeled cholesterol
- CHE (cholesterol efflux) measured using incubated cells and a control. Serum samples from each subject were run in the same assay to eliminate interassay variation.

Independent Variables

- SFA diet
- PUFA diet.

Description of Actual Data Sample:

- *Initial N:* 14 males
- *Attrition (final N):*14 total samples
- *Age:* 18 to 55 years
- *Ethnicity:* Caucasian
- *Anthropometrics:* Body weight and waist circumference measured. Not applicable because of cross-over design
- *Location:* Ethics Committee of the Institute for Clinical and Experimental Medicine approved the study design; however, the actual location of where the study was conducted was not specified.

Summary of Results:

Concentration of Lipids and Lipoproteins, and the Rate of CHE to Serum in 14 Healthy Men at Baseline and After Four Weeks of a High SFA or High PUFA Diet

Variables	Baseline	SFA Diet	PUFA Diet
Total cholesterol (mmol per L)	4.82 (0.86)	5.13 (0.85)	4.65 (0.70)b
Triglycerides (mmol per L)	1.35 (0.7)	1.69 (0.84)	1.64 (0.71)
LDL-cholesterol (mmol per L)	3.24 (0.87)	3.33 (0.69)	3.02 (0.55)b
LDL-cholesterol calc. (mmol per L)	3.01 (0.82)	3.15 (0.65)	2.80 (0.56)b
HDL-cholesterol (mmol per L)	1.19 (0.40)	1.21 (0.30)	1.10 (.032)a
ApoB (g per L)	0.87 (0.33)	0.93 (0.24)	0.90 (0.18)
ApoA-I (g per L)	1.29 (0.15)	1.34 (0.16)	1.25 (0.19)
NEFA (mmol per L)	0.26 (0.18)	0.40 (0.11)	0.43 (0.21)
CHE (%)	10.02 (1.44)	9.74 (1.46)	9.53 (1.41)

a $P<0.05$ (SFA vs. PUFA diet, paired T-test).

b $P<0.01$ (SFA vs. PUFA diet, paired T-test).

Summary

- PUFA diet showed significantly lower concentrations of total cholesterol, LDL-C and HDL-C compared to SFA diet
- Similarly, apoB and ApoA-I concentrations were lower, but not significant (NS)
- Triglycerides and NEFAs were NS different between PUFA and SFA diets
- No change in HDL subfraction distributions were observed between PUFA and SFA diets
- CHE was not different and was comparable to that at baseline
- No correlation found between CHE and lipids and lipoprotein concentrations on both diets

- No correlation between change in CHE and change in HDL-C and apoA-I between diets.

Author Conclusion:

- The decrease in HDL-C resulting from replacement of SFA by PUFA in the diet does not affect the rate of CHE and does not seem to have a detrimental effect
- To the author's knowledge, this study is the first to demonstrate that replacement of SFA by PUFA in humans does not influence CHE to serum from macrophages.

Reviewer Comments:

None.

Research Design and Implementation Criteria Checklist: Primary Research

Relevance Questions		
1.	Would implementing the studied intervention or procedure (if found successful) result in improved outcomes for the patients/clients/population group? (Not Applicable for some epidemiological studies)	Yes
2.	Did the authors study an outcome (dependent variable) or topic that the patients/clients/population group would care about?	Yes
3.	Is the focus of the intervention or procedure (independent variable) or topic of study a common issue of concern to nutrition or dietetics practice?	Yes
4.	Is the intervention or procedure feasible? (NA for some epidemiological studies)	Yes

Validity Questions		
1.	**Was the research question clearly stated?**	Yes
1.1.	Was (were) the specific intervention(s) or procedure(s) [independent variable(s)] identified?	Yes
1.2.	Was (were) the outcome(s) [dependent variable(s)] clearly indicated?	Yes
1.3.	Were the target population and setting specified?	Yes
2.	**Was the selection of study subjects/patients free from bias?**	Yes
2.1.	Were inclusion/exclusion criteria specified (e.g., risk, point in disease progression, diagnostic or prognosis criteria), and with sufficient detail and without omitting criteria critical to the study?	Yes
2.2.	Were criteria applied equally to all study groups?	Yes

2.3.	Were health, demographics, and other characteristics of subjects described?	Yes
2.4.	Were the subjects/patients a representative sample of the relevant population?	Yes
3.	**Were study groups comparable?**	Yes
3.1.	Was the method of assigning subjects/patients to groups described and unbiased? (Method of randomization identified if RCT)	Yes
3.2.	Were distribution of disease status, prognostic factors, and other factors (e.g., demographics) similar across study groups at baseline?	N/A
3.3.	Were concurrent controls used? (Concurrent preferred over historical controls.)	Yes
3.4.	If cohort study or cross-sectional study, were groups comparable on important confounding factors and/or were preexisting differences accounted for by using appropriate adjustments in statistical analysis?	N/A
3.5.	If case control or cross-sectional study, were potential confounding factors comparable for cases and controls? (If case series or trial with subjects serving as own control, this criterion is not applicable. Criterion may not be applicable in some cross-sectional studies.)	N/A
3.6.	If diagnostic test, was there an independent blind comparison with an appropriate reference standard (e.g., "gold standard")?	N/A
4.	**Was method of handling withdrawals described?**	Yes
4.1.	Were follow-up methods described and the same for all groups?	N/A
4.2.	Was the number, characteristics of withdrawals (i.e., dropouts, lost to follow up, attrition rate) and/or response rate (cross-sectional studies) described for each group? (Follow up goal for a strong study is 80%.)	N/A
4.3.	Were all enrolled subjects/patients (in the original sample) accounted for?	Yes
4.4.	Were reasons for withdrawals similar across groups?	N/A
4.5.	If diagnostic test, was decision to perform reference test not dependent on results of test under study?	N/A
5.	**Was blinding used to prevent introduction of bias?**	???
5.1.	In intervention study, were subjects, clinicians/practitioners, and investigators blinded to treatment group, as appropriate?	???
5.2.	Were data collectors blinded for outcomes assessment? (If outcome is measured using an objective test, such as a lab value, this criterion is assumed to be met.)	Yes

5.3.	In cohort study or cross-sectional study, were measurements of outcomes and risk factors blinded?	N/A
5.4.	In case control study, was case definition explicit and case ascertainment not influenced by exposure status?	N/A
5.5.	In diagnostic study, were test results blinded to patient history and other test results?	N/A
6.	**Were intervention/therapeutic regimens/exposure factor or procedure and any comparison(s) described in detail? Were interveningfactors described?**	Yes
6.1.	In RCT or other intervention trial, were protocols described for all regimens studied?	Yes
6.2.	In observational study, were interventions, study settings, and clinicians/provider described?	N/A
6.3.	Was the intensity and duration of the intervention or exposure factor sufficient to produce a meaningful effect?	Yes
6.4.	Was the amount of exposure and, if relevant, subject/patient compliance measured?	Yes
6.5.	Were co-interventions (e.g., ancillary treatments, other therapies) described?	N/A
6.6.	Were extra or unplanned treatments described?	N/A
6.7.	Was the information for 6.4, 6.5, and 6.6 assessed the same way for all groups?	N/A
6.8.	In diagnostic study, were details of test administration and replication sufficient?	N/A
7.	**Were outcomes clearly defined and the measurements valid and reliable?**	Yes
7.1.	Were primary and secondary endpoints described and relevant to the question?	Yes
7.2.	Were nutrition measures appropriate to question and outcomes of concern?	Yes
7.3.	Was the period of follow-up long enough for important outcome(s) to occur?	Yes
7.4.	Were the observations and measurements based on standard, valid, and reliable data collection instruments/tests/procedures?	Yes
7.5.	Was the measurement of effect at an appropriate level of precision?	Yes
7.6.	Were other factors accounted for (measured) that could affect outcomes?	Yes
7.7.	Were the measurements conducted consistently across groups?	Yes
8.	**Was the statistical analysis appropriate for the study design and type of outcome indicators?**	Yes

8.1.	Were statistical analyses adequately described and the results reported appropriately?	Yes
8.2.	Were correct statistical tests used and assumptions of test not violated?	Yes
8.3.	Were statistics reported with levels of significance and/or confidence intervals?	Yes
8.4.	Was "intent to treat" analysis of outcomes done (and as appropriate, was there an analysis of outcomes for those maximally exposed or a dose-response analysis)?	Yes
8.5.	Were adequate adjustments made for effects of confounding factors that might have affected the outcomes (e.g., multivariate analyses)?	Yes
8.6.	Was clinical significance as well as statistical significance reported?	Yes
8.7.	If negative findings, was a power calculation reported to address type 2 error?	N/A
9.	**Are conclusions supported by results with biases and limitations taken into consideration?**	Yes
9.1.	Is there a discussion of findings?	Yes
9.2.	Are biases and study limitations identified and discussed?	Yes
10.	**Is bias due to study's funding or sponsorship unlikely?**	Yes
10.1.	Were sources of funding and investigators' affiliations described?	Yes
10.2.	Was the study free from apparent conflict of interest?	Yes